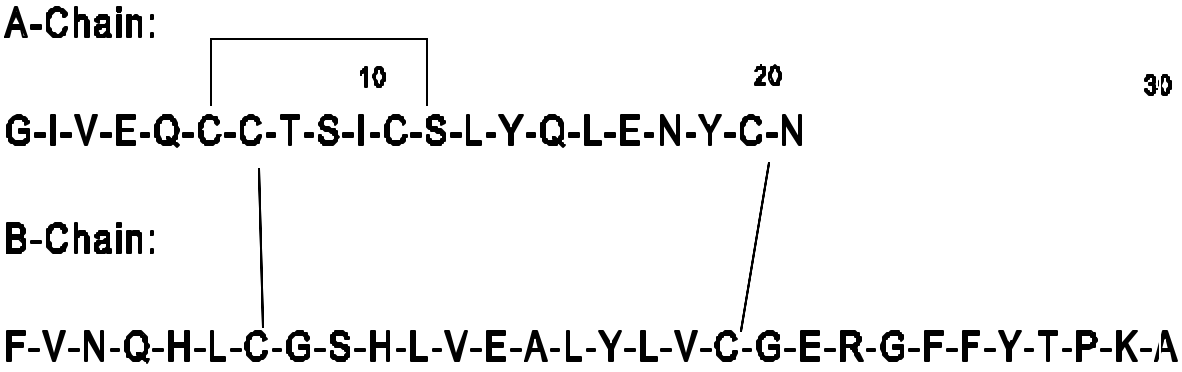
INSULIN from Porcine Pancreas
Sigma Prod. Nos. I5523 and I3505

CAS NUMBER: 2584-58-6



STRUCTURE:

Interchain disulfide bonds are located between the cysteines at positions A7 and B7 and between positions A20 and B19. An intrachain disulfide bond occurs between the cysteines at A6 and A11.[1,2] Porcine insulin differs from human insulin in having alanine in place of threonine at the carboxyl terminal of the B-chain.[1]

PHYSICAL PROPERTIES:
Formula weight: 5777.6
Molecular formula: $C_{256}H_{381}N_{65}O_{76}S_6$
Isoelectric point: pI = 5.3 for the native protein[3]
Sedimentation coefficient ($S_{20,w}$)[4]: 1.95×10^{-13}
Diffusion coefficient ($D_{20,w}$)[4]: 7.30×10^{-7}
Partial specific volume (V_{20})[4]: 0.735 ml/g
Stokes radius (calculated): 11.9 D
E^{M}(278nm) = 6080 (33 mM phosphate, pH 7.0)[5]
$E^{1\%}$(278nm) = 10.6 (33 mM phosphate, pH 7.0)[5]

METHOD OF PREPARATION:
Porcine insulin is purified from pancreas tissue. The final step is crystallization in the presence of zinc. The zinc content is approximately 0.5%. Zinc can be removed by solubilizing insulin in dilute acetic acid, adding excess EDTA to chelate the zinc, and then precipitating the insulin at its pI.[8]

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STABILITY / STORAGE AS SUPPLIED:

Insulin should be stored protected from light in a desiccator at -20°C.

SOLUBILITY / SOLUTION STABILITY:

Insulin is not soluble at neutral pH. It can be solubilized in dilute acetic or hydrochloric acid, pH 2-3. A stock solution can be stored frozen at -20°C in single-use aliquots. Freeze-thaw cycles should be avoided. Alternatively, it can be stored for up to 6 months at 2-8°C if it is sterile filtered through a low protein binding membrane or if it contains a suitable bacteriostat, such as 0.1% thimerosal or sodium azide. Insulin solutions cannot be autoclaved. Insulin can also be solubilized in 125 mM $NaHCO_3$.[6] However, alkaline stock solutions are not recommended, since high pH increases the rate of deamidation and aggregation.[7]

GENERAL REMARKS:

Insulin is produced *in vivo* in the pancreatic ß-cells. The precursor protein (preproinsulin) contains a 23-30 amino acid signal peptide attached to the amino terminal of proinsulin. Proinsulin is composed of the insulin B-chain followed by a connecting peptide (C-peptide) and the A-chain. The signal peptide assists in translocatiing preproinsulin into the lumen of the endoplasmic reticulum, after which it is rapidly cleaved. Proinsulin is then transported to the Golgi complex where it is packaged into granules and converted to insulin. On secretion, equimolar amounts of insulin and C-peptide are released into the blood.[9,10,11]

Potency is determined by HPLC.[2]

Insulin is the primary hormone responsible for controlling the cellular uptake, utilization and storage of glucose, amino acids, and fatty acids while inhibiting the breakdown of glycogen, protein, and fat. Several excellent reviews of the biochemistry, physiology, and pharmacology of insulin have been published.[10,11,12] Additional references can be found in The Merck Index.

REFERENCES:

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NOTES: (continued)

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